

Photosynthetic Comparison of *Carex planostachys* (C₃) and *Aristida purpurea* (C₄) to Light and Water Manipulations

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ABSTRACT

In some southwestern *Juniperus* woodlands, *Carex planostachys* (C₃ sedge) occurs at high cover below the canopy and *Aristida purpurea* (C₄ grass) has high cover in associated grasslands or gaps. Availability of light or water are often suggested causing this variation. Photosynthetic measurements were made as light and water were manipulated in a transplant field study. Monocultures of potted plants were placed below canopy and in an associated gap for three summer months. There were significant main experimental effects on photosynthetic and transpiration rates including location, shade, water level and species. Both species were capable of photosynthetic gain in either location, but CO₂ uptake was three and one-half times greater for the grass in full light. Transpiration rates for both species mostly tracked photosynthetic rates. Examining means plots of three-way interactions showed that no supplemental water had a small effect on *A. purpurea* photosynthetic rates below the canopy, but a large reduction in the gaps. Rates were opposite indicating an interaction. For *C. planostachys* in either location non-watering had limited negative effects. *Aristida purpurea* could carry out photosynthesis at intermediate light levels below the canopy but at reduced rates with no interaction. *Carex planostachys* photosynthesis increased below the canopy when shaded, but decreased without shade in the gaps, an interaction. Water addition impacted photosynthetic rates of both species, but the greatest effect was on *A. purpurea* in the gaps. Light levels and water levels interact to regulate the presence of these two species in *Juniperus* woodlands and associated canopy gaps, they do not act independently. Published on-line www.phytologia.org *Phytologia* 100(1): 91-103 (Mar 16, 2018). ISSN 030319430.

Key Words: A_{net}, E_{leaf}, *Juniperus* woodlands, canopy, gap, drought, sedge, grass

Juniperus communities occur throughout much of the U.S. in semi-arid habitats between the deserts or arid grasslands and mountain forests (Van Auken 2000; Van Auken and McKinley 2008). These communities have 17-80 cm/y rainfall, little summer rain and high rainfall variability with frequent droughts (World Climate 2011). Encroachment of *Juniperus* into many grasslands has created a matrix dominated by woodlands with small associated intercanopy grassland gaps and an overall reduction in grass cover (Reid et al. 1999; West 1999; Archer et al. 2001; Briggs et al. 2002; Van Auken and Smeins 2008). For many communities, the patterns of plant distributions results from the environmental gradients occurring across the habitat, the severity of stressors, and spatial variation in soil type and depth (Givnish 1988; Holmgren et al. 1997; Brady and Weil 2002; Schwinning et al. 2002; Brooker et al. 2008; McDowell et al. 2008; Keddy 2017). Causes for reductions in grass cover following *Juniperus* encroachment are speculative; however, light attenuation by the evergreen *Juniperus* canopy is suspected as a leading agent of change (McPherson et al. 1991). Some plants can adapt to a light regime, thus they are not restricted to a high light or low light environment (Givnish et al. 2004). C₄ grasses typically require full sun in order to maintain high photosynthetic rates (Gehring and Bragg 1992; Knapp and Medina 1999; Turner and Knapp 1996; Wayne and Van Auken 2012; Keddy 2017), but some have a plastic response (Naumburg et al. 2001). Additional controlling abiotic factors could include amount and

seasonality of soil water, temperature and the concentration of other soil nutrients (Wayne and Van Auken 2004; Breshears 2008,) and the factors may act singularly or synergistically to impact plant distributions (Fowler 1990).

During the past 150 – 200 years, *J. ashei* woodlands have encroached throughout much of the central Texas Edwards Plateau. This area was previously mostly a grassland and southern extension of the Great Plains (Diamond and Smeins 1985). In *J. ashei* communities woody cover and grass cover seem inversely related, but the influence of the canopy rapidly declines at the canopy edge (Fuhlendorf et al. 1997; Wayne and Van Auken 2004). Thus, grass cover is high away from the canopy, reduced at the canopy edge and eliminated below the canopy. The idea that *Juniperus* encroachment results in the elimination of most, if not all, herbaceous cover is not always correct and some C₃ sedges increase in cover below the canopy (Gehring and Bragg 1992; Wayne and Van Auken 2004). While the light level below the *J. ashei* canopy is a major controlling influence on the reduced grass cover and biomass below the canopy (Wayne and Van Auken 2004; Young and Bush 2009), a C₃ sedge, *Carex planostachys* (cedar sedge), in many cases has high cover (Wayne and Van Auken 2008). However, at the canopy edge and in the gaps, C₃ and C₄ grasses provide the main cover (Fowler 1990; Wayne and Van Auken 2008).

A second principle factor affecting plant distribution, survival, and physiological function in these arid and semi-arid regions is water availability. In general, drought reduces available water and affects transpiration rates, which has a negative impact on photosynthesis (Tardieu and Simmoneau 1998; Sperry et al. 2003; Keddy 2017), resulting in loss of productivity. Isohydric plants have tight regulation of stomata that limits declines in water stress through reduced transpiration. In severe drought, isohydric plants experience hydraulic loss with infrequent recovery thus limiting their distribution (McDowell 2008). Anisohydric plants have limited stomatal control, are resistant to hydraulic loss and are likely to occur in drought prone areas (Maseda and Fernández 2006; McDowell et al. 2008; McDowell 2011). For example, during drought, some *Juniperus* stems can reach water stress of -10 MPa while maintaining hydraulic conductivity and continued transpiration (Pockman and Sperry 2000; Wayne and Van Auken 2002). Non-woody species typically cavitate (develop gas bubbles in the xylem) at water stress below -3.0 MPa, but maintain some stomatal control to prevent catastrophic water loss (Busch 2001; Sperry et al. 2003; Stiller et al. 2003). C₃ and C₄ grasses may exhibit either isohydric or anisohydric responses, to short-term drought (Tardieu and Simmoneau 1998; McDowell et al. 2008; McDowell 2011).

The causes for distributional differences among grasses and grass like plants in *J. ashei* woodlands are mostly speculative, but are considered related to water and or light availability (Wayne and Van Auken 2012). *Carex planostachys* displays an anisohydric response and appears well adapted to a reduced light environment, but is tolerant of high light levels occurring in gaps (Wayne and Van Auken 2009; 2012). Cover for *C. planostachys* is high in areas below the canopy and at the canopy edge, but at lower cover values in gaps (Wayne and Van Auken 2008). C₄ grasses have high photosynthetic rates in grasslands where canopy imposed light attenuation does not occur (Knapp and Medina 1999; McCarron and Knapp 2001; Fay et al. 2002; Wayne and Van Auken 2012). *Aristida purpurea* occurs in the grasslands associated with *J. ashei* in high light habitats (Fowler 1990; Wayne and Van Auken 2008). Many studies of grass in prairie systems suggest that an overhead canopy is inhibitive to grass growth, either through light attenuation (Fowler 1990; Briggs et al. 2002), allelopathy or limited water (McDowell et al. 2008; Young and Bush 2009; Wayne and Van Auken 2012).

PURPOSE

The purpose of this study was to examine the importance of light and water to the distribution of a C₃ sedge *C. planostachys* found mainly below a *J. ashei* canopy and a C₄ grass *A. purpurea*, found mainly in an associated intercanopy gap.

MATERIALS AND METHODS

Study site and species description

This was a transplant study using monocultures of two species of potted plants to test the influence of light and water on leaf level photosynthesis and transpiration at two locations. The study site (ca. 2000 m²) was located on the University of Texas at San Antonio Texas west campus on the southern edge of the Balcones Escarpment of the Edwards Plateau of central Texas. The area consists of closed canopy *J. ashei* woodlands and associated intercanopy gaps. The woodland vegetation is predominately *J. ashei* and *Quercus virginiana* (Van Auken 1988; Grunstra and Van Auken 2015). The intercanopy gaps include various perennial C₄ grasses and a number of C₃ herbaceous annuals (Terletzky and Van Auken 1996). *Carex planostachys* occurs along a gradient from below the woodland into the gap (Wayne and Van Auken 2008). Supplemental treatments were applied with four combinations of water and shade: water and shade (+S, +W), shade only (+S, -W), water only (-S, +W) and no water and no shade (-S, -W). The species used for this study were *C. planostachys* Kunze (cedar sedge) and *Aristida purpurea* Nutt. Var. *longiseta* (Steud.) Vasey (*Fendler threeawn*) (USDA, NCRS 2017, hereafter *Aristida purpurea* or *Aristida*).

Carex planostachys is a low growing herbaceous perennial with culms 4 – 30 cm long. It is reported mostly in arid areas on thin limestone derived soils of the Edwards Plateau, in north central Texas into Oklahoma, western areas of the Trans-Pecos mountains and south toward the Rio Grande Plains (Correll and Johnston 1979; USDA, NCRS 2017). *Aristida purpurea* is a tufted perennial C₄ grass with culms 15 – 35 cm and leaf blades 4 – 10 cm long. It is reported on rock or sandy slopes of the Trans-Pecos, Edwards Plateau and South Texas plains to North Dakota, Washington State and south into Mexico (Correll and Johnston 1979; USDA, NCRS 2017).

Transplant procedures, site preparation and treatment design

Plastic pots, 15 X 15 cm, were lined with Zip-Loc™ bags and filled with ca. 1400 g of a dried, sieved, low nutrient, Patrick series soils. Individual ramets of *C. planostachys* were obtained on campus below the *J. ashei* woodland in March 2005 and transplanted into prepared pots. Roots were trimmed to 15 cm prior to transplanting. Seeds of *A. purpurea* (Native American Seed, Junction, TX) were planted into additional prepared pots. All pots were initially watered with 500 ml of deionized water and 150 ml of a 25 % fertilizer solution (Miracle-Gro®). To start, pots were kept in a temperature-controlled greenhouse (37° C) and watered as needed to maintain the soil at field capacity.

After germination of *A. purpurea* seeds, plants were thinned to 5 plants per pot. In April 2005, two benches with clear plastic roof panels to exclude rainfall were set up outside the greenhouse to serve as an intermediate site to acclimate plants to outside conditions. In addition, one bench was covered with neutral density shade cloth (75 % occlusion) to reduce light levels (PPF – photosynthetic photon flux) to values similar to those occurring below the *J. ashei* canopy (approximately 700 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, Wayne and Van Auken 2004; Wayne and Van Auken 2008). Light reduction was verified with a LiCor (Lincoln, NE) LI-190 quantum sensor (400-700 nm). All pots were moved outside, with *C. planostachys* on the shaded bench and *A. purpurea* on the unshaded one and provided a second application of 150 ml of a 25 % fertilizer solution. On June 1 2005, pots were randomly assigned to a location and treatment. Prior to establishing pots in the field, the canopy site was cleared of understory woody plants and overstory branches were manipulated with ropes to create a continuous canopy. In the gap, an unshaded site was prepared by removing woody plants and mowing the grass. Pots were given a final application of fertilizer (150 ml of a 25 % solution) and placed into the field.

Below the canopy, individual blocks were placed randomly around a *J. ashei* tree ($n = 5$ trees), between 0.5 and 1.0 m from the trunk. In the gap, blocks were arranged randomly in rows, perpendicular to the solar track, ca. 5 m apart and pots within an individual block were randomly placed ca. 1 m apart.

Supplemental shade was applied with 75 % shade cloth shaped into cones that were 70 cm in height and 50 cm diameter. Shade cones were placed over shade treatment pots and secured to the ground with nails. All pots were checked to ensure a moist surface and marked with labeled flags, either red or blue, for ease in identification of block and treatment. Watering of the no water treatment was discontinued on this day. Plants were monitored every three days and water was added to the water treatment to maintain field capacity, ca. 150 ml for each application.

Physiologic measurements

Photosynthetic measurements were made in late August on fully expanded leaves with a LiCor 6400 portable photosynthetic meter (LiCor, Lincoln, NE) with an integrated light source (measurements made in July, were consistent with August measurements, but not presented). Ambient light levels were determined on a cloudless day to establish global light values for each location with and without supplemental shade. Photosynthetic light was applied with an integrated red-blue LED light source at predetermined global values. Below the canopy, photosynthetic photon flux (PPF) was $700 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, below the canopy with shade PPF was $170 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, in the gaps the PPF was $2000 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and with shade it was $600 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$. Measurements for photosynthesis ($A_{\text{net}} - \mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and transpiration ($E - \text{mol H}_2\text{O}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) were made between 1000 – 1200 hr, after vapor pressure deficit stabilized. Chamber temperature and humidity were held at initial ambient levels, with values established prior to the first measurement. Three to 5 leaves per plant were used, to maximize leaf area, and they were placed into the chamber cuvette and photosynthetic measurements were logged after a stable coefficient of variation ($< 0.2\%$) was reached, approximately 2 to 3 minutes per plant. Because of the time involved for an individual plant response, it was necessary to conduct measurements over two consecutive days.

Analysis

The experimental design was a randomized block design with replications ($n = 5$ blocks), with measurement made in late August. Nested within each block were 2 locations (L). Within each location were 2 species (SP), 2 levels of shade (S) and 2 levels of water (W). Treatments combinations for each species were supplemental shade and supplemental water (S+ W+), supplemental shade and no water (S+ W-), no shade and supplemental water (S- W+) and no shade and no water (S- W-). To test for overall effects, data were analyzed with ANOVA in a mixed model design (SAS 2005) with location nested in blocks, and species, shade and water treated as random effects. For interactions Bonferroni corrections were used to determine where significant differences occurred between location, added shade, added water, and species.

RESULTS

Photosynthesis results for August measurements are presented, but only A_{net} photosynthesis and transpiration E_{leaf} . Net photosynthesis varied significantly by location, shade, water, and species (Table 1), with six significant two-way interactions (S x W, S x SP, W x Sp, L x S, L x W, and L x Sp) and two significant three-way interactions (L x S x W and L x S x Sp). Greatest A_{net} was in the gaps (L), with no shade, but with added water (Figure 1a). *Aristida* had an A_{net} about 3.5 times that of *Carex* (Figure 1a). Transpiration varied significantly by location, shade, and water (Table 1), and there were two significant two-way interactions (L x S and L x W) and two significant three-way interactions (L x S x W and L x S x Sp). Main effects for transpiration were location (highest in gaps), shade (lowest with added shade), and water (greatest when added), with the rate for *Aristida* higher than that of *Carex* (Figure 1b).

For the most part, A_{net} is greatest in the gaps (location) and lower below the canopy, but this is coupled with additional shade having a general suppression of A_{net} which is linked to added water and the species. Because the three-way interactions (L x S x W and L x S x Sp, Table 1) were significant, shade and water effects should be examined and compared simultaneously per species. Thus we used means plots to show the effects of shading (+ and -) and water additions (+ and -) (Figure 2).

For *Aristida*, greatest A_{net} was in the gaps with added water (Figure 2 top-left). Below the canopy when watered, photosynthesis was reduced. Without added water and below the canopy photosynthesis did not change very much, but in the gaps it was reduced to about $6 \mu\text{mol CO}_2 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$. Note that the lines connection the gap treatments and the canopy treatments cross indicating the significant interaction between treatments. When light treatments were examined, the shade+ treatments were lower than the shade- treatments, but the lines for the gap and canopy were parallel indicating no significant interaction.

For *Carex* when water was added canopy and gap photosynthetic values were very similar (Figure 2, top right). When plants were not watered, rates were the same below the canopy or in the gaps, reduced to about $3 \mu\text{mol CO}_2 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$ (Figure 2, top right). When light levels were examined, lowest A_{net} was below the canopy, with added shade (Figure 2, lower right). When shade was removed, *Carex* greatest A_{net} was below the canopy and lowest in the gaps. Note the lines connection the gap and canopy cross indicating a significant interaction.

Transpiration rates are presented in figure 3 and are similar to photosynthetic rates. Transpiration rates for *Aristida* were largest in the gaps when water was added and lowest in the non-watered gap treatment (Figure 3, upper left). Transpiration rates for *Aristida* were also high below the canopy when water was added but lower without added water. Also note the connected gap and canopy lines cross indicating a significant interaction. When shading was examined, transpiration was the same below the canopy or in the gaps (Figure 3, lower left). When shade was removed, transpiration in the gaps was much higher than the rate below the canopy which changed very little, and an interaction is suggested.

For *Carex*, transpiration was highest in the gaps with added water and reduced without addition of water (Figure 3, upper right). Transpiration was lower below the canopy and lowest when water was not added and the connected lines for the gap and the canopy were parallel indicating no interaction. Examining shading for *Carex* showed that highest transpiration was in the gap treatment with no shading and lower in the shade treatment (Figure 3, lower right). When transpiration below the canopy was examined, shade reduced transpiration more than the no shade treatment but the connected lines for the gap and the canopy were parallel indicating no interaction.

DISCUSSION

Plants are found at different points along complex environmental gradients, and the factors that produce these gradients effect plant photosynthetic activity, growth, survival and distribution (McDowell 2011; Keddy 2017). Light and water availability simultaneously affect photosynthetic activity including transpiration or water loss (Chaves et al. 2003; Tuzet et al. 2003). Thus, they are coupled with both influencing carbon gain. Prolonged drought places greater survival risk on plants because of reduced photosynthate to meet metabolic demands (McDowell 2011). During this study light levels and water availability were manipulated below a *J. ashei* canopy and in adjacent intercanopy gaps or locations. Light levels, water availability, and location were significant in the experiment. The manipulation of these factors effected the responses of the two species differently. This led to significant three way interactions in the experiment making interpretation challenging to discern.

Reduced light levels reduced the photosynthetic rate of *Aristida purpurea*, but the canopy or added water moderated the response. This suggested that *Aristida* is a sun plant as reported earlier from measurements of *in situ* plants (Wayne and Van Auken 2004; 2008; 2009). Increased light levels in the gaps reduced the photosynthetic rate of *Carex planostachys* unless shade was added making the gap treatment light levels similar to the canopy light levels. Observations on these plants gradient position when cover and physiologic responses are considered suggest that *C. planostachys* is a shade plant and quite different in its response to light compared to *A. purpurea* (Wayne and Van Auken 2008; 2009; 2012).

Transpiration rates mostly tracked photosynthesis rates, being higher in the gaps, with or without added shade or added water. The *J. ashei* trees were responsible for the additional light attenuation of the canopy. Artificial shade in the gap had a similar effect to that of the woodland canopy. Below the canopy, supplemental shade was additive causing further reductions in carbon uptake in *Aristida*. These results were consistent with the expected response in light availability due to the canopy presence (Muth and Bazzaz 2002; Breshears 2008; Wayne and Van Auken 2012). Distinct gradients in light levels do occur in *J. ashei* woodlands and are related to location, from below the canopy into the gap. While light levels are intermediate at the canopy edge, the focus of this study was the influence of light levels in the closed canopy understory and the gap where the canopy was not present.

Carex planostachys is tolerant of deep shade, with a reported light compensation of ca. $4.2 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and light saturation of ca. $151 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ (Wayne and Van Auken 2009). The results from this study suggest the benefit of light reduction by the canopy to carbon gain by *C. planostachys*. This was apparent both below the canopy with and without supplemental shade and in the gap with supplemental shade. Peak photosynthetic rates were observed between 600 and $700 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, but photosynthetic gain still occurred at reduced light levels ($120 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), which is above the light compensation point (Wayne and Van Auken 2009; 2012).

Aristida purpurea, was expected to have higher photosynthetic rates with increasing light levels because it is a C_4 grass which typically have higher photosynthetic rates than C_3 plants (Keddy 2017). The photosynthetic rate of *A. purpurea* measured in the gaps at high light levels (ca. $2000 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), was expected to be about $20 \mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, but was half that value. Rates were probably lower because plants were in pots with limited soil and water availability. The photosynthetic response of *C. planostachys* was low, typical of shade plants, and what we have reported in the past (Wayne and Van Auken 2009). However, the rate below the canopy with added water suggested soil water reduction and competition with overstory *J. ashei* trees.

Plants can respond to abiotic or biotic factors to conserve water, maintain turgor, prevent catastrophic cavitation, and protect the photosystem (Tyree and Sperry 1989; Galmés et al. 2007; McDowell et al. 2008; McDowell 2011). An increase or decrease in photosynthesis results in a similar response of transpiration. Besides light as a primary regulator of the photosynthetic response, available soil water, temperature, and vapor pressure deficit have an influencing effect (Chaves et al. 2003; Keddy 2017). Despite ca. 60 days of drought in the current study, a reduction in available plant water appeared to have specific impacts on photosynthesis in both species. This is an expected response from anisohydric plants (McDowell et al. 2008; McDowell 2011). During stress photosynthesis should decrease and is related to water stress interfering with metabolic activity (Wong et al. 1979; Sharkey 2005; McDowell 2011). Enhanced transpiration is reported to occur in many plants while under severe stress, despite stomatal control, and provides some benefit through evaporative cooling, reducing leaf temperature to protect from damaging heat (Larcher 1995; Valladares and Pearcy 1997; Chaves et al. 2003). *Aristida purpurea*, a C_4 plant may avoid drought stress by maintaining smaller stomatal apertures reducing water loss (McCarron and Knapp 2001).

Differences between species photosynthetic responses were observed and were related to the light and water levels manipulated during the experiment. Other species have demonstrated similar responses (McDowell et al. 2008; McDowell 2011). In terms of functional groups, *C. planostachys* appears like a shade tolerant plant and *A. purpurea* as a shade intolerant or sun plant (Wayne and Van Auken 2009, 2012). This in part explains the high cover of *C. planostachys* reported below the *J. ashei* canopy and the high cover *A. purpurea* in adjacent gaps (Wayne and Van Auken 2004; 2008; 2009). Temperature differences between treatment locations, thought slight and not examined here, could help further delineate why the two species are found in different though adjacent habitats (Sharkey 2005). However, both species appeared to gain carbon in intermediate light habitats, which helps explain near equal cover

of both species at the *J. ashei* canopy edge (Wayne and Van Auken 2008). Canopy moderation of environmental factors, favoring various herbaceous plants and woody seedlings has been reported for deciduous woodlands and some evergreen woodland (Hughes et al. 2006). In prairie systems, tall grasses have a similar effect to that of a woodland canopy reducing the stress on short C₃ and C₄ grass (McCarron and Knapp 2001; Fay et al. 2002; Fay et al. 2003).

LITERATURE CITED

- Archer, S.R., Boutton, T.W. and Hibbard, K.A. 2001. Trees in grasslands: Biogeochemical consequences of woody plant expansion, in E.D. Schulze, et al. (eds.). Global Biogeochemical Cycles in the Climate System. Academic Press, NY..
- Brady, N. and Weil, R. 2002. The nature and properties of soils, 13th edition. Prentice Hall, NY.
- Breshears, D.D. 2008 Structure and function of woodland mosaics: consequences of patch-scale heterogeneity and connectivity along the grassland-forest continuum. in O.W. Van Auken, (ed.). Western North American *Juniperus* communities: a dynamic vegetation type. Springer, NY.
- Briggs, J.M., Hoch, G.A. and Johnson, L.C. 2002. Assessing the rate, mechanisms and consequences of the conversion of tallgrass prairie to *Juniperus virginiana* forest. Ecosystems 5:578-586.
- Brooker, R.W., et al. 2008. Facilitation in plant communities: the past, the present, and the future. Journal of Ecology 96:18-34.
- Busch, J. 2001. Characteristic values of key ecophysiological parameters in the genus *Carex*. Flora 196:405-430.
- Chaves, M.M. and Maroco, J.P. and Pereira, J.S. 2003. Understanding plant responses to drought - from genes to whole plant. Functional Plant Biology 30:239-264.
- Correll, D.S. and Johnston, M.C. 1979. Manual of the vascular plants of Texas. Texas Research Foundation, Renner, TX.
- Diamond, D.D. and Smeins, F.E. 1985. Composition, classification, and species response patterns of remnant tallgrass prairies in Texas. American Midland Naturalist 113:294-308.
- Fay, P.A., et al. 2002. Altered rainfall patterns, gas exchange, and growth in grasses and forbs. International Journal of Plant Sciences 163:549-557.
- Fay, P.A., Carlise, J.D., Knapp, A.K., Blair, J.M. and Collins, S.L. 2003. Productivity responses to altered rainfall patterns in a C₄-dominated grassland. Oecologia 137:245-251.
- Fowler, N.L. 1990. The effects of competition and environmental heterogeneity on three coexisting grasses. Journal of Ecology 78:389-402.
- Fuhlendorf, S.D., Smeins, F.E. and Taylor, C.A. 1997. Browsing and tree size influences on ashe juniper understory. Journal of Range Management 50:507-512.
- Galmés, J., Flexas, J., Savé, R. and Medrano, H. 2007 Water relations and stomatal characteristics of Mediterranean plants with different growth forms and leaf habits: responses to water stress and recovery. Plant Soil 290:139-155.
- Gehring, J.L. and Bragg, T.B. 1992. Changes in prairie vegetation under eastern redcedar (*Juniperus virginiana* L.) in an eastern Nebraska bluestem prairie. American Midland Naturalist 128:209-217.
- Givnish, T.J. 1988. Adaptation to sun and shade: a whole-plant perspective. Australian Journal of Plant Physiology 15:63-92.
- Givnish, T.J., Montgomery, R.A. and Goldstein, G. 2004. Adaptive radiation of photosynthetic physiology in the Hawaiian Lobeliads: light regimes, static-light responses, and whole-plant compensation points. American Journal of Botany 91:228-246.
- Grunstra, M.B. and Van Auken, O.W. 2015. Photosynthetic characteristics of *Garrya ovata* Benth. (Lindheimer's silktassle, Garryaceae) at ambient and elevated levels of light, CO₂ and temperature. Phytologia 97:103-119.
- Holmgren, M., Scheffer, M. and Huston, M.A. 1997. The interplay of facilitation and competition in plant communities. Ecology 78:1966-1975.

- Hughes, R.F., Archer, S., Asner, G.P., Wessman, C.A., McMurtry, C., Nelson, J., and Ansley, R.J. 2006. Changes in aboveground primary production and carbon and nitrogen pools accompanying woody plant encroachment in a temperate savanna. *Global Change Biology* 12:1733-1747.
- Knapp, A.K. and Medina, E. 1999. Successes of C₄ photosynthesis in the field: lessons from communities dominated by C₄ plants, in R. Sage and Monson, R. (eds.). *The Biology of C₄ Plants*. Academic Press, NY.
- Keddy, P.A. 2017. *Plant Ecology: origins, processes, consequences*. Second ed. Cambridge University Press, NY.
- Larcher, W. 1995. *Physiological plant ecology: ecophysiology and stress physiology of functional groups* 3rd edition. Springer-Verlag, NY.
- Maseda, P.H. and Fernández, R.J. 2006. Stay wet or else: three ways in which plants can hydraulically adjust to their environment. *Journal of Experimental Botany* 57:3963-3977.
- McCarron, J.K. and Knapp, A.K. 2001. C₃ woody plant expansion in a C₄ grassland: are grasses and shrubs functionally distinct. *American Journal of Botany* 88:1818-1823.
- McDowell, N., et al. 2008. Mechanisms of plant survival during drought: why do some plants survive while others succumb to drought? *New Phytologist* 178:719-739.
- McDowell, N. G. 2011. Mechanisms linking drought, hydraulics, carbon metabolism, and vegetation mortality. *Plant Physiology* 155: 1051-1059.
- McPherson, G.R., Rasmussen, G.A., Wester, D.B. and Masters, R.A. 1991. Vegetation and soil zonation patterns around *Juniperus pinchotii* plants. *Great Basin Naturalist* 51:316-324.
- Muth, C.C. and Bazzaz, F.A. 2002. Tree canopy displacement at forest gap edges. *Canadian Journal of Forest Research* 32:247-254.
- Naumburg, E., De Wald, L.E. and Kolb, T.E. 2001. Shade responses of five grasses native to southwestern U.S. *Pinus ponderosa* forests. *Canadian Journal of Botany* 79:1001-1009.
- Pockman, W. T. and Sperry, J. S. 2000. Vulnerability to xylem cavitation and the distribution of Sonoran Desert vegetation. *American Journal of Botany* 87: 1287-1299.
- Reid, K.D., Wilcox, B.P., Breshears, D.D. and MacDonald, L, 1999. Runoff and erosion in a piñon-juniper woodland: influence of vegetation patches. *Soil Science Society of America Journal* 63:1869-1879.
- SAS. 2005. *SAS/STAT 9.1 User's Guide*. SAS Publishing, Cary, NC.
- Schwinning, S., Davis, K., Richardson, L. and Ehleringer, J.R. 2002. Deuterium enriched irrigation indicates different forms of rain use in shrub/grass species of the Colorado Plateau. *Oecologia* 130:345-355.
- Sharkey, T.D. 2005. Effects of moderate heat stress on photosynthesis: importance of thylakoid reactions, rubisco deactivation, reactive oxygen species, and thermotolerance provided by isoprene. *Plant, Cell and Environment* 26:269-277.
- Sperry, J.S., Stiller, V. and Hacke, U.G. 2003. Xylem hydraulics and the soil-plant-atmosphere continuum: opportunities and unresolved issues. *Agronomy Journal* 95:1362-1370.
- Stiller, V., Lafitte, H.R. and Sperry, J.S. 2003. Hydraulic properties of rice and the response of gas exchange to water stress. *Plant Physiology* 132:1698-1706.
- Tardieu, F. and Simmoneau, T. 1998. Variability among species of stomatal control under fluctuating soil water status and evaporative demand: Modelling isohydric and anisohydric behaviors. *Journal of Experimental Biology* 49:419-432.
- Terletzky, P.A. and Van Auken, O.W. 1996. Comparison of cedar glades and associated woodlands of the southern Edwards Plateau. *Texas Journal of Science* 48:55-67.
- Turner, C.L. and Knapp, A.K. 1996. Responses of a C₄ grass and three C₃ forbs to variation in nitrogen and light in tallgrass prairie. *Ecology* 77:1738-1749.
- Tuzet, A., Perrier, A. and Leuning, R. 2003. A coupled model of stomatal conductance, photosynthesis and transpiration. *Plant, Cell and Environment* 26:1097-1116.
- Tyree, M.T. and Sperry, J.S. 1989. The vulnerability of xylem to cavitation and embolism. *Annual Review of Plant Physiology and Plant Molecular Biology* 40:19-38.

- USDA, NRCS. 2017. The PLANTS Database (<http://plants.usda.gov>, 16 October 2017). National Plant Data Team, Greensboro, NC 27401-4901 USA.
- Valladares, F. and Pearcy, R.W. 1997. Interactions between water stress, sun-shade acclimation, heat tolerance and photoinhibition in the sclerophyll *Heteromeles arbutifolia*. Plant, Cell and Environment 20:25-36.
- Van Auken, O.W. 1988. Woody vegetation of the southeastern escarpment and plateau. in B.B. Amos and Gehlbach, F.R. (eds.). Edward's Plateau vegetation. Baylor Univ. Press, Waco, TX.
- Van Auken, O.W. 2000. Shrub invasion of North American semiarid grasslands. Annual Review of Ecology and Systematics 31:197-215.
- Van Auken, O.W. and McKinley, D.C. 2008. Western North American *Juniperus* communities: patterns and causes of distribution and abundance. in O.W. Van Auken, (ed.). Western North American *Juniperus* communities: a dynamic vegetation type. Springer, NY.
- Van Auken, O.W. and Smeins, F.E. 2008. Western North American *Juniperus* communities: patterns and causes of distribution and abundance. in O.W. Van Auken. (ed.). Western North American *Juniperus* communities: a dynamic vegetation type. Springer, NY.
- Wayne, R. and Van Auken, O.W. 2002. Spatial and temporal patterns of xylem water potential in *Juniperus ashei* seedlings. Southwestern Association of Naturalists 47: 153-161.
- Wayne, E.R. and Van Auken, O.W. 2004. Spatial and temporal abiotic changes along a canopy to intercanopy gradient in Central Texas *Juniperus ashei* woodlands. Texas Journal of Science 56:35-54.
- Wayne, E.R. and Van Auken, O.W. 2008. Comparisons of the understory vegetation of *Juniperus* woodlands. in O.W. Van Auken. (ed.). Western North American *Juniperus* communities: a dynamic vegetation type. Springer, NY.
- Wayne, E.R. and Van Auken, O.W. 2009. Light responses of *Carex planostachys* from various microsites in a *Juniperus* community. Journal of Arid Environments 73:435-443.
- Wayne, E.R. and Van Auken, O.W. 2012. Differences in gas exchange rates provide insight into distributions of C3 sedges and C4 grass in central Texas Savanna. Phytologia 94: 71-90.
- West, N.E. 1999. Juniper-Piñon savannas and woodland of western North America. in R.C. Anderson, Fralish, J.S. and Baskin, J.M. (eds.). Savannas, barrens, and rock outcrop plant communities of North America. Cambridge University Press, NY.
- Wong, S.C., Cowan, I.R. and Farquhar, G.D. 1979. Stomatal conductance correlates with photosynthetic capacity. Nature 282:424-426.
- World Climate. 2011. Climate Data for 29°N 98°W. Accessed April 1, 2016.
<<http://www.worldclimate.com/cgi-bin/grid.pl?gr=N29W099>>
- Young, G.P. and Bush, J.K. 2009. Assessment of the allelopathic potential of *Juniperus ashei* on germination and growth of *Bouteloua curtipendula*. Journal of Chemical Ecology 35:74-80.

Table 1. ANOVA table testing for within significant main effects of location (L), shade (S), water (W), species (Sp) and all interactions. Parameters measured were leaf net photosynthesis ($A_{\text{net}} - \mu\text{mol CO}_2 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) and transpiration ($E_{\text{leaf}} - \text{mmol H}_2\text{O} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$). Measurements were made in late August 2005. *P*-values are given for effects included in the model. ns = not significant.

August	$A_{\text{net}}\text{-}P$	$E_{\text{leaf}}\text{-}P$
L	≤ 0.0001	≤ 0.0001
S	≤ 0.0001	< 0.05
W	≤ 0.0001	< 0.01
Sp	≤ 0.0001	ns
S x W	≤ 0.0001	ns
S x Sp	≤ 0.0001	ns
W X Sp	≤ 0.0001	ns
L x S	≤ 0.0001	≤ 0.0001
L x W	≤ 0.0001	≤ 0.0001
L x Sp	≤ 0.0001	≤ 0.0001
L x S x W	≤ 0.0001	≤ 0.0001
L x S x Sp	≤ 0.0001	≤ 0.0001
L x W x Sp	ns	ns
S x W x Sp	ns	ns
L x S x W x Sp	ns	ns

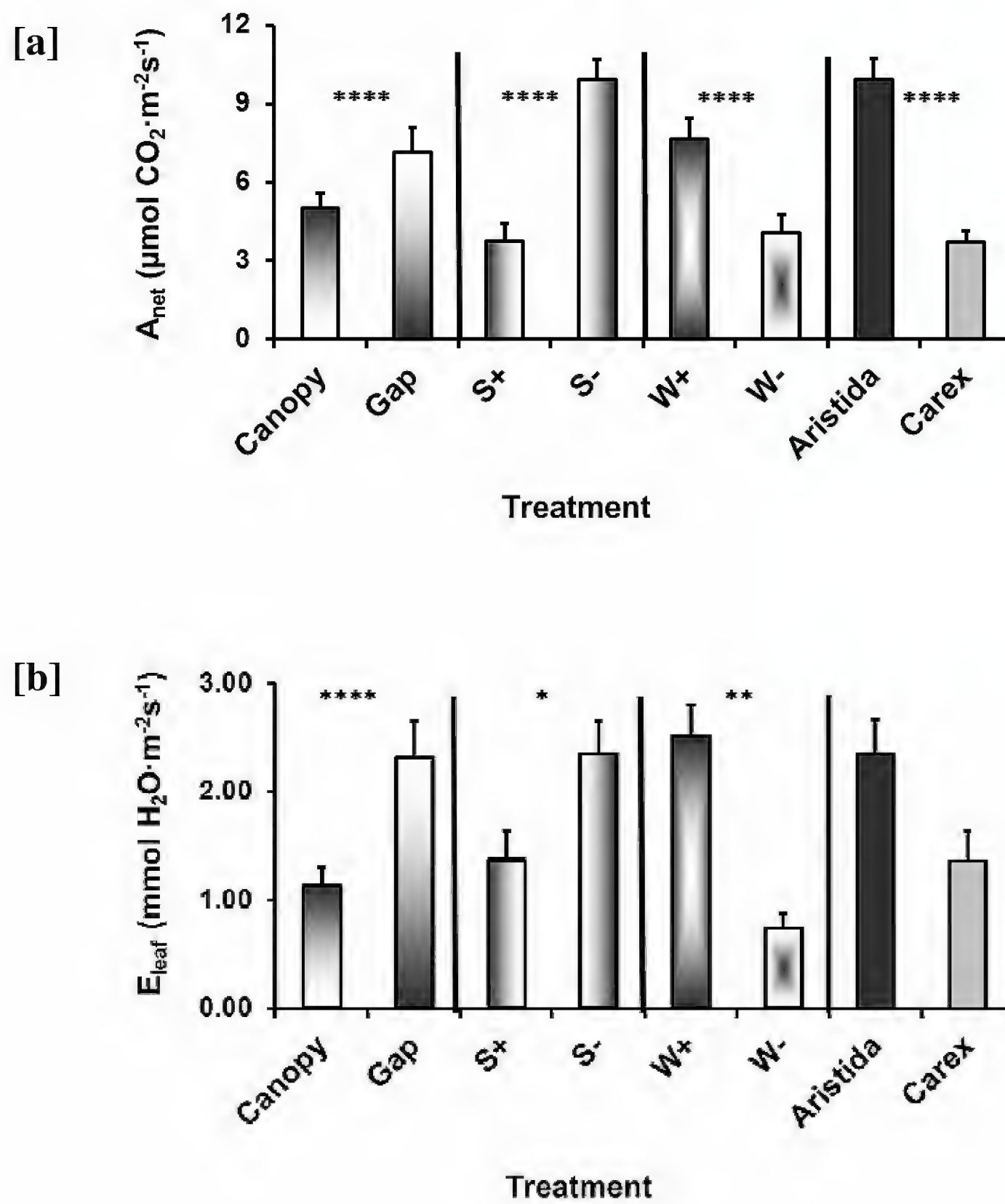


Figure 1. Mean main effect plots ($\bar{x} \pm SE$) for [a, upper]-net photosynthesis (A_{net}) and [b, lower]-transpiration (E_{leaf}) in August. Treatments tested were for the effects of location (Canopy or Gap), shade (S+ or S-), water (W+ or W-) and species (*Aristida* or *Carex*). Vertical bars separate treatment groups. Treatment groups with symbols above indicate significant main effects. * = $P < 0.05$, ** = $P < 0.01$ and **** = $P \leq 0.0001$.

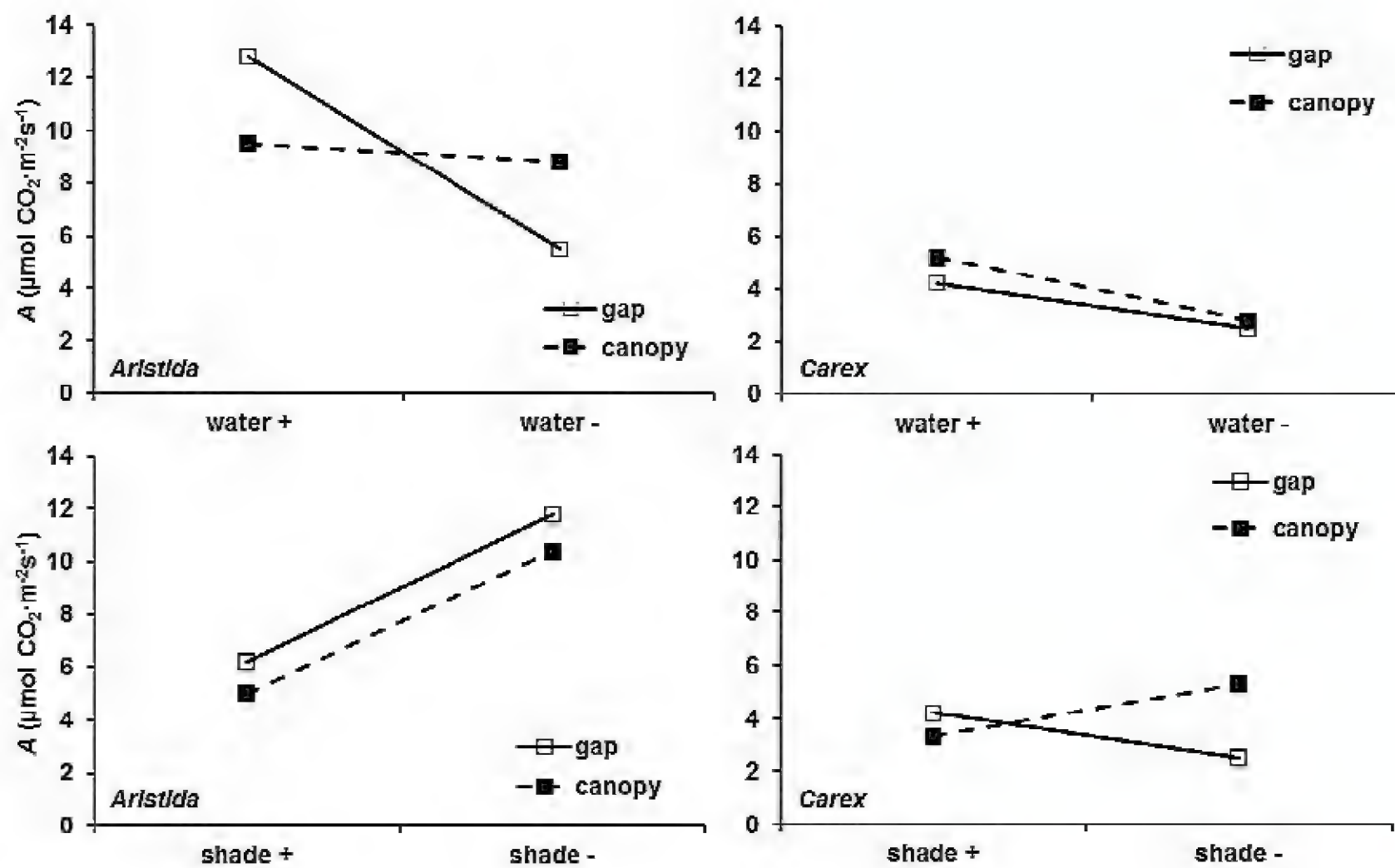


Figure 2. Means plots of the shade \times water interaction on net photosynthesis (A_{net}) for *Aristida longiseta* (left side) and *Carex planostachys* (right side) in August. Measurements presented are treatment average on plants occurring below the *J. ashei* canopy and in an associated intercanopy gap. Treatments were supplemental water and no added water (W+ and W-) upper plots and supplemental shade and no added shade (S+ and S-) lower plots.

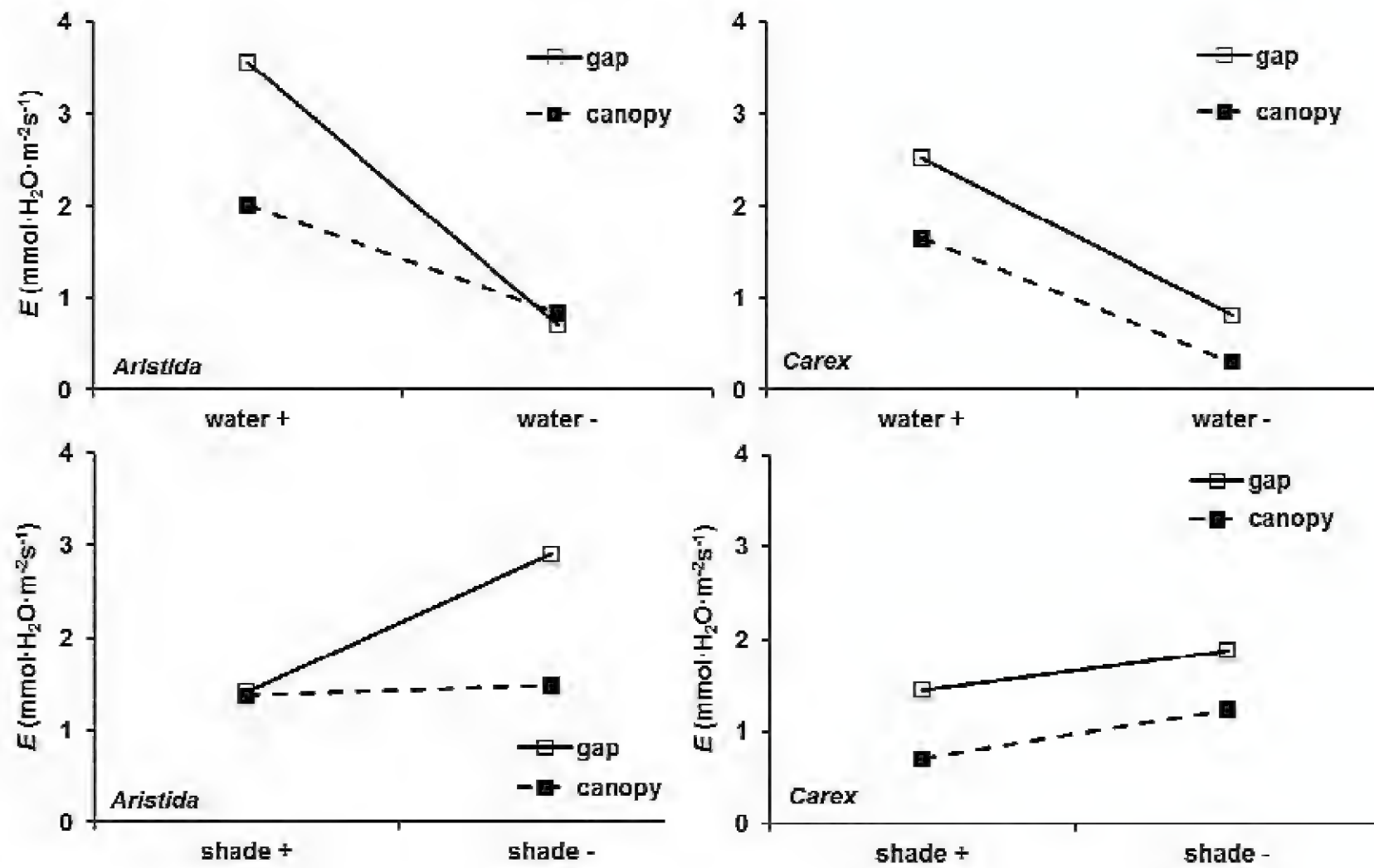


Figure 3. Means plots of the shade \times water interaction on leaf transpiration (E_{leaf}) for *Aristia longiseta* and *Carex planostachys* in August. Measurements presented are treatment average on plants occurring below the *J. ashei* canopy and in an associated intercanopy gap. Treatments were supplemental water and no added water (W+ and W-) upper plots and supplemental shade and no added shade (S+ and S-) lower plots.